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AVAILABILITY OF NIACIN IN CORN AND MILO FOR SWINE¹

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NIACIN has been recognized for several years as a dietary essential for swine. Its presence in feedstuffs is well known, but the biological availability of niacin in certain cereal grains commonly fed to swine has not been clearly established. Research has been reported indicating the niacin of white corn and wheat is largely unavailable to the pig (Kodicek *et al.*, 1956; Kodicek *et al.*, 1959 and Luce *et al.*, 1966). Several workers have also obtained evidence that the niacin of corn is in a bound form and largely unavailable to the rat. (Laguna and Carpenter, 1951; Pearson *et al.*, 1957; Harper *et al.*, 1958; Kodicek and Wilson, 1959; Squibb *et al.*, 1959 and Kodicek, 1960).

No information could be found in the literature on the availability of niacin in yellow corn and milo for swine, therefore these studies were conducted.

Experimental Procedure

Experiment 1. Twenty-four cross bred barrows which averaged 20.3 kg. of body weight were allotted at random to six replications of four treatments. The experiment was conducted for 70 days. The pigs were housed and individually fed in concrete-floored pens which were equipped with self-feeders and waterers. The experimental unit was completely enclosed and was equipped with a thermostatically-controlled heating system. Temperature in this unit was maintained at approximately 20° C.

Composition of the experimental rations is presented in table 1. The purified ration which assayed 1.85 mg. niacin per kg. was fed with and without a supplement of 17.6 mg. of niacin per kg. of feed. The corn ration which contained 13.5 mg. of total niacin per kg. of feed was also fed with and without a supplement of 17.6 mg. of crystalline niacin

per kg. of feed. Vitamin-free casein and gelatin were used as protein supplements to maintain the total protein content of all rations at approximately 16%. The calculated tryptophan content was held constant (0.14%) in all treatments.

The growth study was terminated on the 58th day of the experiment and a metabolism trial was conducted on the surviving 19 pigs. Total collections of feces and urine were made for 3 days after a 3-day adjustment period.

Experiment 2. Sixteen Yorkshire-Hampshire crossbred barrows which averaged 37.0 kg. of bodyweight were allotted at random within litters to four treatments. The experiment was conducted for 33 days. The pigs were housed and group-fed in concrete-floored pens which were equipped with self-feeders and waterers. The experimental unit was of the open-shed type and the temperature fluctuated with the environmental temperature.

Composition of the experimental diets is shown in table 1. The diets were formulated with 40, 60, or 80% yellow corn. The total niacin content of these rations supplied exclusively by corn was 8.11, 12.17 and 16.22 mg. per kg., respectively. One additional ration was fed which contained 80% yellow corn with 8.11 mg. of crystalline niacin added per kg. of feed. Corn starch was used to keep the energy levels of all rations approximately the same. Gelatin and vitamin-free casein were used as supplements to maintain all rations at approximately 14% protein. The calculated tryptophan level was held constant (0.11%) in all treatments.

All pigs were fed the 40% corn ration for 10 days. At the end of the initial period, 12 pigs were chosen at random within litters from the 16 pigs on trial and placed in metabolism cages for daily collections of feces and urine for 5 days after a 3-day adjustment period. Upon the completion of the initial metabolism trial the 12 pigs were randomly allotted to the 40% corn, 60% corn, 80% corn and 80% corn with added niacin. The pigs were fed these rations for 10 days

¹ Published with the approval of the Director as paper No. 1772, Journal Series, Nebraska Agricultural Experiment Station.

² Department of Animal Science. Acknowledgement is made to John Welch for assistance in development of certain of the laboratory procedures and to P. F. Cunningham and associates for care of the experimental animals.

and were then returned to the metabolism cages for a second metabolism trial lasting 5 days after a 3-day adjustment period.

Experiment 3. Ten Hampshire-Yorkshire crossbred barrows which averaged 32.6 kg. of bodyweight were allotted to two treatments. The experiment was conducted for 41 days. Housing and management were the same as described in Experiment 2.

The experimental rations and their composition are shown in table 1. Rations fed were 80% milo with and without a supplement of 33.07 mg. of crystalline niacin per kg. of feed. Vitamin-free casein and gelatin were used as supplements to maintain all rations at approximately 16.0% protein. The calculated tryptophan level was kept constant (0.11%) in both treatments.

The pigs were fed their respective rations for 21 days and then 3 pigs from each ration treatment were chosen at random and placed in metabolism cages. Total collections were made for 3 days after a 3-day preliminary period. At the end of this metabolism trial, the two rations were reversed, i.e., the pigs previously fed the 80% milo ration were

switched to the 80% milo ration plus niacin and the pigs fed the 80% milo ration plus niacin were switched to the 80% milo ration. Total collections were made again for 3 days after a 3-day adjustment period. Blood samples were obtained from each pig at the end of the trials and processed as described by Luce *et al.* (1966).

Metabolism cages, collection procedures and sample processing for all experiments were similar to those described previously (Luce *et al.*, 1966). Nitrogen, N'-methylnicotinamide (NMN), N'-methyl-2-pyridone-5-carboxamide (2 PY), niacin, diphosphopyridine nucleotide (DPN) and hematocrits were determined by procedures outlined previously by the above authors.

Student's *t* test as outlined by Steel and Torrie (1960) was used to determine differences between treatment means. A probability level of $P < .05$ or less was accepted as being significant. In Experiments 2 and 3, treatment differences for average daily feed intake and feed required per kilogram of gain could not be tested since the animals were grouped and the treatments not replicated.

TABLE 1. COMPOSITION OF EXPERIMENTAL RATIONS

Ration designation	Experiment 1		Experiment 2			Experiment 3
	A,B ^a	C,D ^b	A	B	C,D ^c	A,B ^a
Treatment	Purified	Corn	40% Corn	60% Corn	80% Corn	80% Milo
Ingredients, %						
Ground yellow corn	79.8	40.0	60.0	80.0
Ground milo	80.0
Corn starch	71.2	35.7	17.9	0.2	1.4
Casein	11.7	3.5	6.7	5.9	4.2	4.1
Gelatin	6.1	6.1	4.9	3.7	3.3	3.3
Sucrose	2.5	2.5	4.0	4.0	4.0	4.0
Lard	2.5	2.5	4.0	4.0	4.0	3.0
Salt (iodized)	0.5	0.5	0.5	0.5	0.5	0.5
Monosodium phosphate	2.6	1.8	0.4	0.2
Dicalcium phosphate	2.7	2.7	2.7	2.6
Ground limestone	1.8	1.7
Trace minerals ^e	0.1	0.1	0.1	0.1	0.1	0.1
Vitamin premix ^f	1.0	1.0	1.0	1.0	1.0	1.0
L-lysine	0.5
% Protein, Chemical analysis	16.38	16.78	14.62	14.81	14.75	16.61
% Tryptophan, calculated	0.14	0.14	0.11	0.11	0.11	0.11
Niacin, (mg./kg.) (total)						
microbiological assay	1.85	13.21	8.11	12.17	16.22	31.22
% Calcium, calculated	0.68	0.66	0.71	0.71	0.71	0.70
% Phosphorus, calculated	0.66	0.66	0.70	0.70	0.70	0.72

^a Supplemented with 11.35 mg. of niacin per kg. of feed for ration B.

^b Supplemented with 23.81 mg. of niacin per kg. of feed for ration C.

^c Supplemented with 8.11 mg. of niacin per kg. of feed for ration D.

^d Supplemented with 33.07 mg. of niacin per kg. of feed for ration B.

^e Calcium Carbonate Company, Quincy, Ill. Standard swine mix. Content in %: Fe, 10; Mn, 10; Cu, 1.0; Co, 0.10; I, 0.30; Zn, 10 and Ca, 9.1.

^f Contributed sufficient vitamins to bring the total content to the following levels per kg. of complete ration: vitamin A, 4409.0 I.U.; vitamin D₂, 397.0 I.U.; riboflavin, 5.5 mg.; pantothenic acid, 17.6 mg.; choline, 881.8 mg.; vitamin B₁₂, 15.4 mcg.

TABLE 2. EFFECT OF SOURCE OF NIACIN ON AVERAGE DAILY GAINS, DAILY FEED INTAKE AND FEED REQUIRED PER KG. OF GAIN^a (EXPERIMENT 1)

Ration designation	A	B	C	D
Treatment	Purified	Purified+niacin	Corn	Corn+niacin
Niacin level, mg./kg. (total)	1.85	13.21	13.51	37.32
Pigs started per treatment	6	6	6	6
Pigs finished per treatment	2 ^b	5 ^b	6	6
Av. initial wt., kg.	22.7	21.2	20.3	20.3
Av. daily gain, kg.	0.03	0.13	0.55 ^c	0.73 ^c
Av. daily feed intake, kg.	0.54	0.69	1.54 ^c	1.92 ^c
Feed per kg. gain, kg.	20.10	7.16	2.80	2.65

^a Duration of experiment was 70 days.^b Pigs either died or became paralyzed and were removed from treatment.^c Rations C and D differ significantly ($P<.01$).

Results

Experiment 1. The results of the growth study are presented in table 2. Pigs fed the purified niacin-deficient ration performed very poorly and developed rather typical symptoms of a niacin deficiency within 2 or 3 weeks. The symptoms observed were diarrhea, rough hair coat, dermatitis, stiffness and paralysis of the hind quarters, irritability, anorexia and severe retardation in rate of gain. Two of the six pigs fed this ration died during the third week of the experiment and two more were destroyed when they became paralyzed and unable to rise. Two survived the experiment.

A slight response in pig performance was obtained by the addition of niacin to the purified ration. When measured by weight gain, the response was considered to be poor. Apparently the purified ration used in this study was not readily acceptable to the pigs as evidenced by their low daily feed intake which was apparent from the start. The pigs fed the purified ration plus niacin also showed symptoms of retarded rate of gain and anorexia, but the symptoms were less severe. One of the pigs fed this ration also became

paralyzed and was removed from the experiment. No niacin deficiency symptoms were observed in the pigs fed the corn ration with the exception of a reduced rate of gain.

The average daily gain of 0.73 kg. for the pigs fed the corn ration plus niacin was significantly greater ($P<.01$) than the 0.55 kg. made by those fed the corn ration. The average daily feed intake was higher ($P<.01$) and feed required per kg. of gain was less for the pigs fed the corn ration plus niacin than for those fed the corn ration.

Values for urinary excretion of NMN and 2 PY, nitrogen digestibility and nitrogen retention are shown in table 3. Average daily urinary excretion of NMN and 2 PY by the pigs was increased greatly when niacin was added to the corn ration. NMN and 2 PY excretion were increased from 0.86 to 3.49 mg./per day ($P<.01$) and from 0.51 to 4.46 mg./per day respectively, by the addition of niacin to the 80% corn ration. Pigs fed the purified ration plus niacin also excreted greater amounts of NMN and 2 PY than the pigs fed the purified ration.

Although all pigs were observed to be excreting NMN, the averages for 2 PY ex-

TABLE 3. EFFECT OF SOURCE OF NIACIN ON URINARY NMN AND 2 PY EXCRETION, NITROGEN DIGESTIBILITY AND NITROGEN RETENTION (EXPERIMENT 1)

Ration designation	A	B	C	D
Treatment	Purified	Purified+niacin	Corn	Corn+niacin
Niacin level, mg./kg. (total)	1.85	13.21	13.51	37.32
Av. daily niacin intake, mg.	0.17	4.31	20.73	58.44
Av. daily NMN excretion, mg.	0.58	0.71	0.86 ^a	3.50 ^a
Av. daily 2PY excretion, mg.	0.76	1.04	0.51 ^a	4.46 ^a
Av. daily nitrogen intake, gm.	2.34	8.79	42.15	44.94
Av. daily nitrogen digestion, %	90.6	94.6	89.1	88.9
Av. daily nitrogen retention, % of intake	-96.4	14.7	45.6	35.0

^a Rations C and D differ significantly ($P<.01$).

cretion include one pig fed the purified ration, one pig fed the purified ration plus niacin and two pigs fed the corn ration in which no 2 PY could be detected in the urine.

Nitrogen digestibility was 90.6 and 94.6%, respectively, for the pigs fed the purified ration and the purified ration plus niacin. Nitrogen digestibility for the pigs fed the corn ration and the corn ration plus niacin was 89.1 and 88.9%, respectively, and did not differ significantly. The pigs fed the niacin-deficient, purified ration excreted more nitrogen than they consumed, whereas the pigs fed the purified ration plus niacin showed an average daily retention of 14.7% of nitrogen intake. The percent of nitrogen intake

pigs fed the 40% corn ration than was required by those fed the other treatments.

The effect of rations and ration changes on urinary excretion of NMN for the experimental periods are shown in figure 1. NMN excretion increased in a linear manner for the pigs fed the 40, 60 and 80% corn rations without supplemental niacin and the 80% corn ration with supplemental niacin, but the linear component was not statistically significant.

A linear increase was observed in NMN excreted daily when measured 13 days after the pigs were switched from the 40% corn ration (control) to the 60 and 80% corn rations and 80% corn ration with added

TABLE 4. EFFECT OF SOURCE OF NIACIN ON AVERAGE DAILY GAINS, FEED INTAKE, FEED CONVERSION, NIACIN DIGESTIBILITY AND NITROGEN DIGESTIBILITY AND RETENTION^a (EXPERIMENT 2)

Ration designation	A	B	C	D
Treatment	40% Corn	60% Corn	80% Corn	80% Corn + niacin
Niacin level, mg./kg. (total)	8.10	12.16	16.21	24.32
Pigs per treatment	3	3	3	3
Av. initial wt., kg.	42.4	43.4	42.2	43.3
Av. daily gain, kg.	0.33	0.45	0.38	0.53
Feed per kg. gain, kg.	3.67	2.54	2.97	2.54
Metabolism study				
Av. daily niacin intake, mg.	10.50	15.42	21.15	40.18
Niacin digestibility, %	75.9	71.5	77.3	70.3
Av. daily nitrogen intake, gm.	31.4	30.1	30.8	39.6
Av. daily nitrogen digestibility, %	91.5	89.0	91.2	86.5
Av. daily nitrogen retention, % of intake	53.7	55.3	47.2	48.9

^a Duration of experiment was 33 days.

retained by the pigs fed the corn ration was greater than for those fed the corn ration plus niacin (45.6 versus 35.0%).

Experiment 2. The results of the growth study and the niacin digestibility values observed are presented in table 4. Average daily gains for the pigs fed the 40, 60 and 80% corn rations without supplemental niacin did not differ significantly. Average daily gains of the pigs fed the 80% corn ration with supplemental niacin were higher (0.53 kg. per day) but were not significantly different from the gains of the pigs fed the 80% corn ration without supplemental niacin. Average daily feed intake was similar for the pigs fed the 40, 60 and 80% corn rations without supplemental niacin. The pigs fed the 80% corn ration plus niacin consumed more feed than those fed the other rations. Considerably more feed was required per kg. of gain for the

niacin. Nevertheless, values observed were not significantly different.

Only a trace of 2 PY could be detected in the urine of three animals; one each on the 60% corn, 80% corn and 80% corn plus niacin rations.

A summary of nitrogen digestibility and retention data is also shown in table 4. No significant differences were noted in nitrogen digestibility or percent of nitrogen intake retained.

Experiment 3. The results of the growth study are presented in table 5. Pigs fed the milo ration supplemented with 33.1 mg. of niacin per kg. of feed gained 0.07 kg. more per day than those fed the unsupplemented ration. The difference in gains was not statistically significant. The average daily feed intake was also higher for the pigs fed the milo ration plus niacin (2.58 vs. 2.36 kg.).

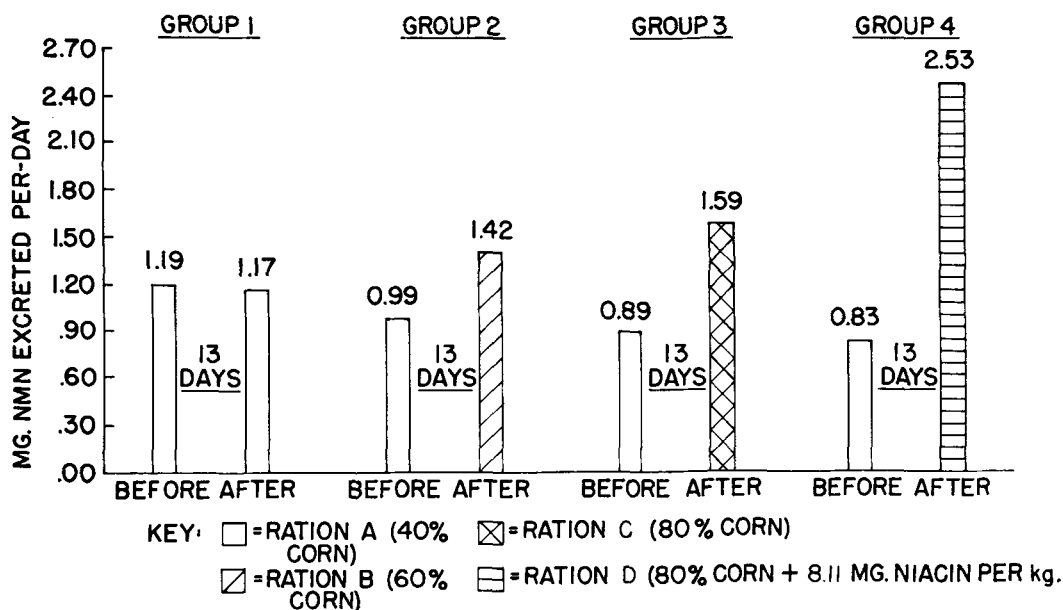


Figure 1. Effect of rations (before) and ration changes (after) on urinary excretion of NMN by swine (Exp. 2).

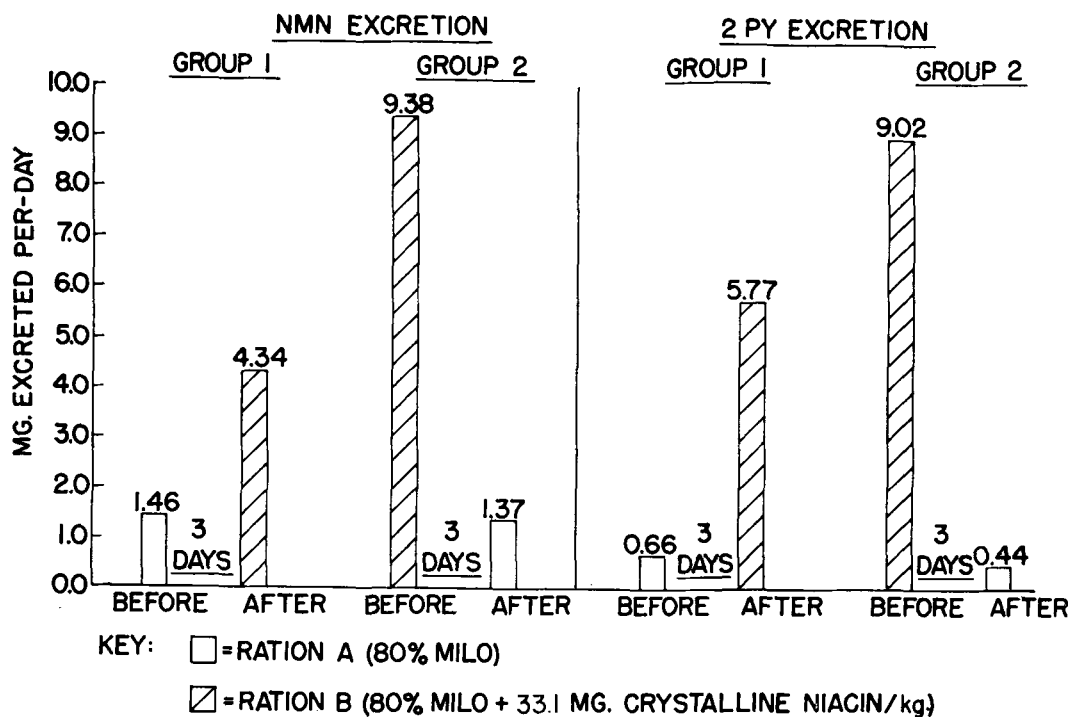


Figure 2. Effect of ration (before) and ration reversal (after) on urinary excretion of NMN and 2 PY by swine (Exp. 3).

Feed per kg. of gain was the same for both treatments.

The effect of ration and ration reversal on urinary excretion of NMN and 2 PY is shown in figure 2. The treatments had a marked influence on NMN and 2 PY excretion by the pigs. The average daily NMN excretion for the pigs fed the milo ration plus niacin was significantly greater ($P < .01$) than the excretion observed for the pigs fed

+5.11 and -8.58 mg., respectively. After ration reversal, NMN excretion was significantly changed ($P < .01$).

The effect of ration and ration reversal on blood DPN levels is shown in figure 3. Blood DPN levels were higher for the pigs fed the milo ration plus niacin than for the pigs fed the milo ration (23.9 vs. 18.6 mcg. per ml. of erythrocytes). Seven days after ration reversals, the pigs switched from the ration to

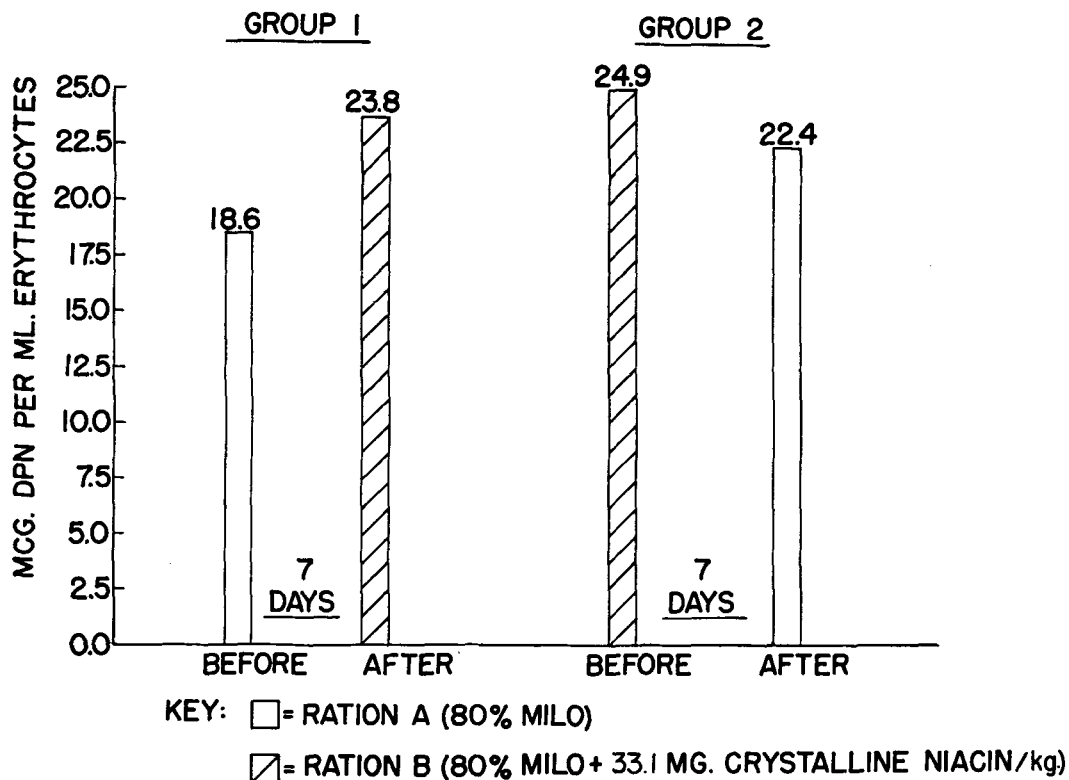


Figure 3. Effect of ration (before) and ration reversal (after) on blood DPN levels in swine (Exp. 3).

the milo ration. Similarly, the average daily 2 PY excretion of the pigs fed the milo ration plus niacin was greater than the excretion by the pigs fed the milo ration.

Three days after ration reversals, the pigs switched from the milo ration to the milo ration plus niacin showed an increase of 2.88 mg. per day in excretion of NMN. Pigs switched from the ration plus niacin to the milo ration showed a decrease of 8.01 mg. in average daily NMN excretion. Differences observed in 3 days in average daily 2 PY excretion for the same ration reversals were

the ration plus niacin showed an increase of 5.3 mcg. in average DPN levels per ml. of erythrocytes, whereas the pigs switched from the milo ration plus niacin to the milo ration showed a decrease of 2.5 mcg. However, the differences observed between rations before and after reversals were not statistically significant.

Nitrogen digestibility and retention data are also summarized in table 5. No significant differences were noted in nitrogen digestibility or percent of nitrogen intake retained for the pigs fed the two experimental rations.

TABLE 5. EFFECT OF SOURCE OF NIACIN ON AVERAGE DAILY GAIN, FEED INTAKE AND FEED REQUIRED PER KG. OF GAIN, EXCRETION OF NIACIN AND NITROGEN DIGESTIBILITY AND RETENTION^a (EXPERIMENT 3)

Ration designation	A	B
Treatment	80% milo	80% milo + niacin
Niacin level, mg./kg. (total)	31.3	64.4
Pigs per treatment	5	5
Av. initial wt., kg.	32.7	32.5
Av. daily gain, kg.	0.73	0.80
Feed per kg. gain, kg.	3.22	3.22
Metabolism study ^b		
Av. daily niacin intake, kg	72.42	127.99
Av. daily niacin digestibility, %	80.9 ^c	89.8
Niacin excreted in urine, % of intake	6.2 ^c	3.5 ^c
Av. daily nitrogen intake, gm.	59.4	58.4
Av. daily nitrogen digestibility, %	71.0	71.1
Av. daily nitrogen retention, % of intake	53.0	50.3

^a Duration of experiment was 41 days.

^b One pig removed from ration B because of failure to eat.

^c Rations A and B differ ($P < 0.05$) for niacin excreted in urine.

Discussion

The results of these studies indicate that the addition of crystalline niacin improved weight gains of growing pigs fed rations containing corn or milo as the primary source of energy. The increased gains observed in the pigs by the addition of niacin to the corn rations are in agreement with results reported by several workers (Braude *et al.*, 1946; Luecke *et al.*, 1947, Luecke *et al.*, 1948 and Becker and co-workers, 1963). The total niacin content of the corn or milo ration used in these studies equalled or exceeded the level recommended by the National Research Council (1964) as the requirement for growing pigs. Therefore our results indicate that the niacin in yellow corn and milo is not appreciably available to swine.

Urinary NMN and 2 PY excretion were observed to be valid indices of the niacin intake of swine. Generally, NMN and 2 PY were observed to increase in a linear manner as levels of niacin were increased in the rations. The increased excretion of NMN or 2 PY in the urine of pigs by the addition of niacin to the diet agrees with the findings of several workers (Luecke *et al.*, 1947; Luecke *et al.*, 1948; Perlzweig *et al.*, 1950 and Self *et al.*, 1960). Also, it was observed

that great changes could be produced in NMN and 2 PY excretion within 3 days by the addition or removal of niacin from the diet of swine. This indicates the immediate effect which niacin intake has on these urinary metabolites.

NMN excretions appeared to be less variable than 2 PY excretion among pigs fed the same ration. In fact, 2 PY could not be detected constantly in the urine of pigs fed a low-niacin diet. This observation is in agreement with the work of Perlzweig *et al.* (1950) who were not able to detect 2 PY in the urine of one pig on a stock ration (ingredients not given). The failure of 2 PY to appear constantly in the urine was explained by Brown *et al.* (1958) who suggested that 2 PY was only excreted in significant amounts in humans when niacin or tryptophan is present in the diet in excess of metabolic needs.

Crystalline niacin when fed to pigs produced much greater excretions of NMN and 2 PY than the niacin originating from corn and milo. This observation was also shown by Luce *et al.* (1966) with wheat diets for swine. The data did suggest that the niacin (at least in corn) was not totally unavailable as increased corn levels in the diet of pigs caused slight increases in NMN excretion (if even only a low percent were available then one would expect a linear increase in NMN excretion with increased levels of corn).

Urinary excretion of free niacin by swine did not seem to adequately reflect differences in niacin intake. Only slight increases in free niacin excretion were observed by large increases of niacin in the diet of swine. This observation has also been shown by Luecke *et al.* (1947) and Wintrobe *et al.* (1945).

The niacin digestibility values (70–89%) obtained in both the corn and milo trials are similar to the average value of 77% reported for humans by Denko *et al.* (1946). The high digestibility values indicate that the niacin of cereal grains is highly digestible, and is destroyed in the intestinal tract or experiences some other unknown fate. Thus niacin digestibility probably does not present a true picture of the availability of niacin ingested by the pig.

The increased blood DPN levels which were observed in pigs when niacin was added to the ration clearly demonstrate the effect of this vitamin on pyridine nucleotide synthesis. Similar relationships have been shown in rats and humans by several workers (Kohn and

Klein, 1939; Hoagland and Ward, 1942; Handler and Kohn, 1943, Hoagland and others, 1943; Duncan and Sarett, 1951; Burch *et al.*, 1955 and Morrison *et al.*, 1963). The changes observed in 7 days in blood DPN levels by adding or removing niacin from the diet also demonstrated the rapid effect which this vitamin has on the DPN concentration of the blood. Duncan and Sarett (1951) reported that the administration of niacin to humans caused a rise in blood DPN levels within 4 hours.

Tryptophan has been shown by numerous workers to be a precursor of niacin in animals and microorganisms. It has been shown also to be a substitute for niacin in pigs by Luecke *et al.* (1948) and Powick *et al.* (1948). Nevertheless, tryptophan conversion to niacin in swine is apparently a very inefficient process (50 to 1) as shown by Firth and Johnson (1956).

Reber *et al.* (1951) demonstrated that a tryptophan deficiency produced a negative nitrogen balance in swine. An attempt was made in all experiments to keep the tryptophan level at or above 0.11% recommended by Hays (1961) as being the minimum requirement of swine of this weight range. Since no general reduction in nitrogen retention was observed in the pigs fed the low-niacin diets, in all probability no appreciable tryptophan conversion to niacin occurred.

Summary

Three experiments were conducted with 46 crossbred barrows to determine the availability of niacin in yellow corn and milo for growing pigs. Urinary excretion of free niacin, N'-methylnicotinamide (NMN), N'-methyl-2-pyridone-5-carboxamide (2 PY), digestibility of niacin, blood levels of diphosphopyridine nucleotide (DPN), rate of gain, feed conversion and digestibility and retention of nitrogen were used as criteria for determining availability. In the first experiment, average daily gains and NMN excretion were increased significantly ($P < .01$), by the addition of 17.6 mg. of niacin per kg. of feed to a corn ration. In the second experiment, average daily gains were similar for pigs fed 40, 60 or 80% corn ration without added niacin. Gains were increased by the addition of 8.1 mg. of crystalline niacin per kg. of feed to the 80% corn ration. NMN excretion increased in a linear manner for pigs fed the 40, 60 and 80% corn rations and the 80%

corn ration with added niacin. In the third experiment, average daily gains, NMN and 2 PY excretions and blood DPN levels were increased by the addition of 33.0 mg. of niacin per kg. to an 80% milo ration. Differences observed in NMN excretion were significant ($P < .025$). Generally only slight differences were noted in free niacin excretion, nitrogen retention and digestibility and feed conversion. The results indicate that the niacin of corn and milo is largely unavailable to swine.

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